

CLAIMS

1. (currently amended) A method for ~~obtaining purified native~~ purifying Interleukin-4 (IL-4) or muteins thereof comprising: (a) expressing the IL-4 or muteins thereof in a prokaryotic cell thereby forming inclusion bodies containing IL-4 or muteins thereof in said prokaryotic cell; (b) disrupting the prokaryotic cell to release the inclusion bodies; (c) washing separating the inclusion bodies in a washing buffer capable of solubilizing lipids bound to the surface of the inclusion bodies or contained in cell wall fragments from the cell debris; (d) solubilizing the inclusion bodies in a solution that includes a denaturing agent comprising a guanidinium salt, thereby obtaining a guanidine-denatured ~~denaturing the~~ IL-4 or muteins thereof; (e) purifying separating the guanidine-denatured ~~denatured~~ IL-4 or muteins thereof using an immobilized metal chelate affinity chromatography (IMAC) system; and (f) releasing the IL-4 or muteins thereof from the IMAC system; and (g) renaturing the guanidine-denatured IL-4 or muteins thereof in the presence of an artificial chaperone, thereby obtaining ~~the~~ purified native IL-4 or muteins thereof, ~~wherein the step of separating the denatured IL-4 or muteins thereof with the IMAC system provides an average recovery of the IL-4 or muteins thereof of better than 80% and a purity of the IL-4 or muteins thereof of about 90% as estimated by SDS-PAGE analysis.~~

2. (canceled)

3. (previously presented) The method according to claim 2, wherein the washing buffer comprises a non-ionic detergent, an ionic surfactant or a zwitterionic detergent.

4. (previously presented) The method according to claim 2, wherein the washing buffer is a buffer which maintains the pH between 7 and 10.
5. (previously presented) The method according to claim 2, wherein the washing buffer additionally contains a chelating substance.
6. (previously presented) The method according claim 5, wherein the chelating substance is selected from the group consisting of ethylenediaminetetraacetic acid (EDTA), ethyleneglycol-O,O' bis-(2-aminoethyl)-N,N,N',N'-tetraacetic acid (EGTA), nitriloacetic acid (NTA) and trans-1,2-diamino-cyclohexan-N,N,N',N'-tetraacetic acid (CDTA).
7. (previously presented) The method according to claim 1, wherein the Interleukin-4 is IL-4 R121D Y124D.
- 8-9. (canceled)
10. (currently amended) The method according to claim 1 9, wherein the artificial chaperone is a cyclic dextrin or linear dextrin.
11. (previously presented) The method according to claim 1, wherein the prokaryotic host is *E. coli*.

12. (previously presented) The method according to claim 1, wherein the IL-4 is mIL-4 Q116D Y119D.

13-18. (canceled)

19. (previously presented) The method according to claim 3, wherein the zwitterionic detergent is selected from the group consisting of CHAPS, CHAPSO, desoxycholate and the zwittergent series (N-alkyl-N,N-ditnethyl-3-ammonio-1-propanesulfonate).

20. (canceled)

21. (new) The method according to claim 1, further comprising the step of releasing the IL-4 or muteins thereof from the IMAC system prior to renaturing the IL-4 or muteins thereof.

22. (new) The method according to claim 1, further comprising the step of releasing the IL-4 or muteins thereof from the IMAC system subsequent to renaturing the IL-4 or muteins thereof.